Bioorganic & Medicinal Chemistry Letters 18 (2008) 5729–5733

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

N-Glycine-sulfonamides as potent dual orexin 1/orexin 2 receptor antagonists

Hamed Aissaoui [a,*], Ralf Koberstein [a], Cornelia Zumbrunn [a], John Gatfield [a], Catherine Brisbare-Roch [b], Francois Jenck [b], Alexander Treiber [c], Christoph Boss [a]

[a] Drug Discovery Chemistry and Biochemistry, Actelion Pharmaceuticals Ltd, Gewerbestrasse 16, 4123 Allschwil, Basel-Land, Switzerland
[b] Drug Discovery Pharmacology, Actelion Pharmaceuticals Ltd, Gewerbestrasse 16, 4123 Allschwil, Basel-Land, Switzerland
[c] Drug Discovery Drug Metabolism, Actelion Pharmaceuticals Ltd, Gewerbestrasse 16, 4123 Allschwil, Basel-Land, Switzerland

ARTICLE INFO

Article history:
Received 4 September 2008
Revised 22 September 2008
Accepted 23 September 2008
Available online 26 September 2008

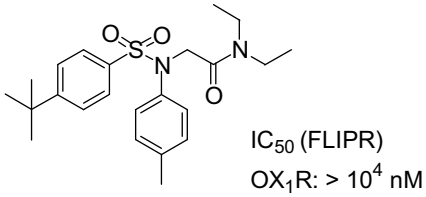
Keywords:
Orexin
Dual orexin antagonist
N-Glycine-sulfonamide
Oral bioavailability
Brain penetration

ABSTRACT

A series of dual OX_1R/OX_2R orexin antagonists was prepared based on a *N*-glycine-sulfonamide core. SAR studies of a screening hit led to compounds with low nanomolar affinity for both receptors and good oral bioavailability. One of these compounds, **47**, has demonstrated in vivo activity in rats following oral administration.

The orexins (orexin A and orexin B, also named hypocretin 1 and 2, respectively) are hypothalamic peptides discovered in 1998 by two independent research groups.[1,2] These two neuropeptides play an important role in the regulation of the sleep–wake cycle and related hypothalamic functions. They are endogenous ligands of two G protein-coupled receptors, namely, OX_1R and the OX_2R and are proteolytically derived from the same precursor peptide.[1,2] While both neuropeptides bind with similar affinity to the OX_2R, orexin A binds with slightly higher affinity to OX_1R than orexin B.[2] Patients with narcolepsy have degenerated hypothalamic orexin neurons and low levels of orexins in cerebrospinal fluid.[3] Activation of orexin neurons contributes to the promotion and maintenance of wakefulness, and conversely, relative inactivity of orexin neurons allows the onset of sleep.[4] Consequently, blocking orexin signaling with receptor antagonists may provide a new mechanism for decreasing wakefulness and, thus, a novel therapeutic opportunity for the treatment of insomnia. Recently, we have reported that a dual OX_1R/OX_2R receptor antagonist (ACT-078573, almorexant) elicited somnolence without inducing cataplexy in rats, dogs, and humans.[5] Herein, we describe a novel series of orexin receptor antagonists based on a *N*-glycine-sulfonamide core. Compounds with excellent potency on both receptors and suitable pharmacokinetic behavior were found.

High-throughput screening of our chemical libraries against a CHO cell line expressing the human orexin receptors using a FLIPR (fluorometric imaging plate reader)-based calcium assay[6] identified the achiral and low molecular weight aryl containing *N*-glycine-sulfonamide **1** (2-[(4-*tert*-butyl-benzenesulfonyl)-*p*-tolyl-amino]-*N*,*N*-diethyl-acetamide) as a potent and selective OX_2R antagonist with very poor oral bioavailability ($F = 1\%$) in Wistar rats.



1

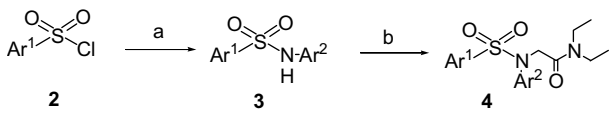
IC_{50} (FLIPR)
OX_1R: > 10^4 nM
OX_2R: 5 nM
F = 1%

To improve potency on both receptors, preliminary structure–activity relationship (SAR) studies around **1** started by keeping constant the *N*,*N*-diethyl-sulfonyl-acetamide moiety and by exploring structural modifications of the two aryl rings.

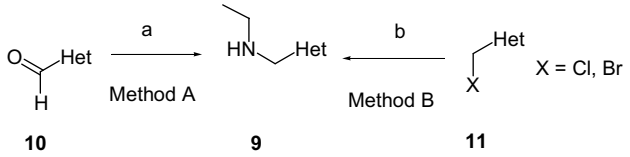
Many of the *N*-glycine-sulfonamide derivatives evaluated were prepared according to the procedure outlined in Scheme 1. The

* Corresponding author. Tel.: +41 61 565 65 63; fax: +41 61 565 65 00.
E-mail addresses: aissaoui.hamed@actelion.com, ahmed.aissaoui@tiscali.fr (H. Aissaoui).

0960-894X/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2008.09.079



Scheme 1. General synthesis of *N*-glycine-sulfonamide derivatives (route A). Reagents and conditions: (a) Ar_2–NH_2, pyridine, rt, 70–85%; (b) 2-chloro-*N*,*N*-diethyl-acetamide, K_2CO_3, DMF, 80 °C, 70–80%.



Scheme 3. General synthesis of ethyl heterocyclic-methyl amines. Reagents and conditions: (a) 2 M $EtNH_2$ in THF, $NaBH_4$, MeOH, rt, 12–63%; (b) 2 M $EtNH_2$ in THF, rt, 24–98%.

appropriate aryl-sulfonyl chlorides **2** were coupled with an aniline derivative in the presence of pyridine. The resulting sulfonamide intermediates **3** with a diverse range of functionality provided the desired final compounds **4** by reaction with 2-chloro-*N*,*N*-diethyl-acetamide in the presence of K_2CO_3.

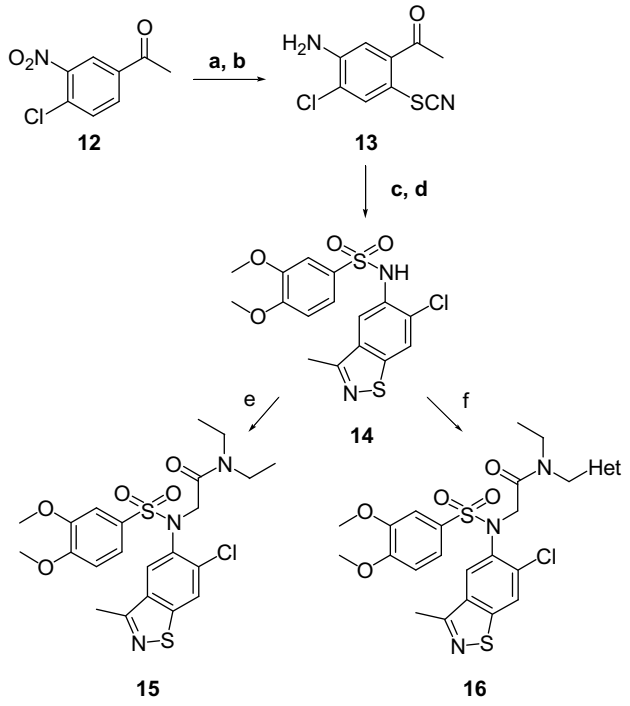
For compounds where one ethyl group was replaced with a heterocycle-CH_2- unit, a modified route was pursued with 3,4-dimethoxybenzenesulfonyl chloride **5** (Scheme 2). Coupling with the appropriate aniline in the presence of pyridine followed by reaction with *tert*-butyl bromoacetate in the presence of K_2CO_3 furnished the sulfonamide intermediate **7**. Removal of the *tert*-butyl ester under acidic conditions (TFA) in the presence of triethylsilane, followed by amide bond formation (i.e., PyBOP, *i*-Pr_2NEt) with an ethyl heterocyclic-methyl amine derivative **9** provided final compounds **8**.

The ethyl heterocyclic-methyl amine derivatives **9** were prepared by reductive amination of the appropriate aldehyde **10** with ethylamine in the presence of sodium borohydride (method A) or by coupling of the suitable heterocyclic halide **11** with ethylamine (method B) (Scheme 3).

The synthesis of compounds whereby Ar^2 is a substituted benzisothiazole moiety, is depicted in Scheme 4.

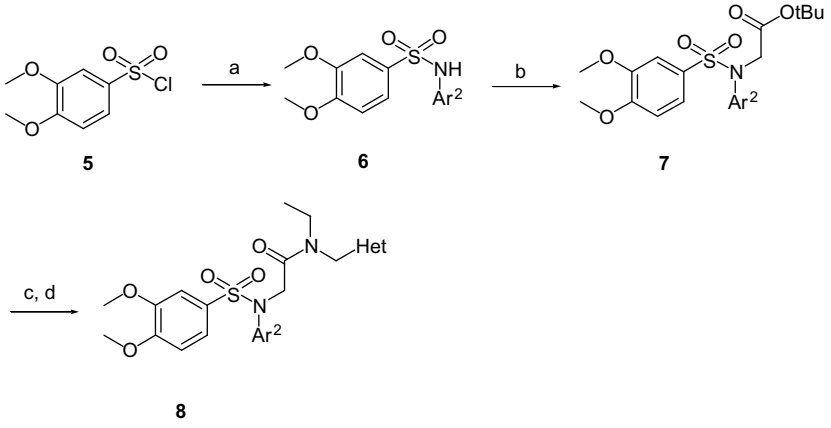
The commercially available acetophenone **12** was reduced with $SnCl_2{\cdot}2H_2O$ in the presence of concentrated HCl in refluxing MeOH. Subsequent bromination in AcOH followed by reaction with sodium thiocyanate (NaSCN) afforded the thiocyanato derivative **13**.[7] Cyclization with concentrated ammonia[7] followed by reaction with 3,4-dimethoxybenzenesulfonyl chloride in the presence of pyridine furnished the sulfonamide intermediate **14.** Reaction with 2-chloro-*N*,*N*-diethyl-acetamide in the presence of K_2CO_3 afforded the target compound **15.** Additional analogs containing the heterocycle-CH_2 unit, **16,** were prepared in the same manner as described in Scheme 2.

Replacing the 4-*tert*-butylbenzene moiety of **1** by a less lipophilic substituent such as a 3,4-dimethoxybenzene ring, did not change the potency or selectivity profiles as exemplified with compound **17** (Table 1). Introduction of further electron-donating



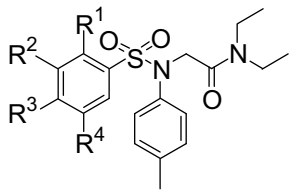
Scheme 4. General synthesis of the substituted benzisothiazole derivatives. Reagents and conditions: (a) $SnCl_2{\cdot}2H_2O$, 37% HCl, MeOH, reflux, 100%; (b) NaSCN, Br_2, AcOH, rt, 99%; (c) 25% NH_4OH, rt, 52%; (d) 3,4-dimethoxybenzenesulfonyl chloride, pyridine, rt, 87%; (e) 2-chloro-*N*,*N*-diethyl-acetamide, K_2CO_3, DMF, 80 °C, 66%; (f) i—*tert*-butyl bromoacetate, K_2CO_3, DMF, 80 °C, rt, 80–87%; (ii) TFA, Et_3SiH, DCM, rt, 90–95%; (iii) 9, PyBOP, *i*-Pr_2NEt, DMF, rt, 30–53%.

substituents such as methyl, ethyl, or methoxy groups resulted in a significant drop in potency towards the OX_2R, especially for



Scheme 2. General synthesis of *N*-glycine-sulfonamide derivatives (route B). Reagents and conditions: (a) Ar_2–NH_2, pyridine, rt, 70–85%; (b) *tert*-butyl bromoacetate, K_2CO_3, DMF, 80 °C, rt, 90–97%; (c) TFA, Et_3SiH, DCM, rt, 90–95%; (d) 9, PyBOP, *i*-Pr_2NEt, DMF, rt, 50–70%.

Table 1
SAR studies on Ar^1 moiety



Compound	R^1	R^2	R^3	R^4	hOX_1R[a]	hOX_2R[a]
1	H	H	*t*-Bu	H	>10,000	5
17	H	OMe	OMe	H	>10,000	19
18	H	H	Et	H	>10,000	89
19	H	H	Me	H	>10,000	232
20	H	H	OMe	H	>10,000	661
21	H	H	Br	H	>10,000	142
22	H	H	F	H	>10,000	346
23	H	H	CN	H	>10,000	1187
24	H	H	OCF_3	H	>10,000	1822
25	Me	H	H	H	>10,000	58
26	Cl	H	H	H	>10,000	800
27	F	H	H	H	>10,000	3372
28	H	Me	H	H	>10,000	60
29	H	Cl	H	H	>10,000	139
30	H	F	H	H	>10,000	839

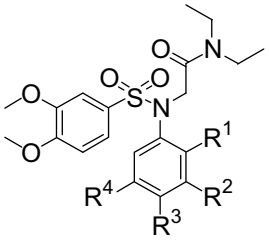
[a] IC_{50} values in nM (FLIPR assay).

compound **20**. The presence of an electron-withdrawing substituent in the *para*-position such as a halogen atom or a cyano group led to less potent, selective OX_2R antagonists as illustrated with compounds **21**–**24**. Similar biological results were obtained with *ortho*- and *meta*-substituted derivatives as exemplified by compounds **25**–**30**.

In a second effort to improve OX_1R potency, we have decided to explore a variety of substitution patterns with different electronic properties at one or more positions of the anilinic phenyl ring in compound **2** (Table 2).

As shown in Table 2, it is apparent that the presence of an electron-withdrawing substituent (preferably a chlorine atom) in the *ortho*-position (R^1) and an electron-donating group such as a dimethylamino group in the *meta*-position (R^4) was very important to obtain significantly improved affinity for the OX_1R (compounds **31** and **32**). On the contrary, introducing only electron-donating or electron-withdrawing substituents in the 2,5-positions resulted in a dramatic loss of potency on both orexin receptors (compounds **33**–**35**). The 2,3-disubstituted and 2,4-disubstituted derivatives were rather OX_2R-selective antagonists (compounds **36**–**38**) with compound **38** being one of the most potent OX_2R antagonists found to date. Inversion of the electronic properties in the 2,5-disubstitution pattern led to compounds with a substantial drop of affinity for the OX_1R but only a small loss of activity towards the OX_2R (compound **39**). Similar tendencies were observed when the dimethylamino group was replaced either by a methyl or an isopropyl group (compounds **40** and **41**). Further structural modifications of the dimethylamino group by elongation (compounds

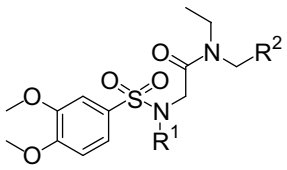
Table 2
SAR studies on Ar^2 moiety

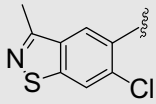
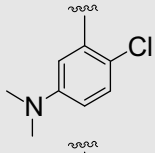
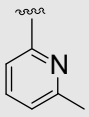
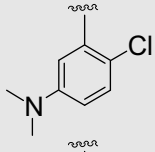
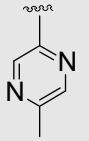
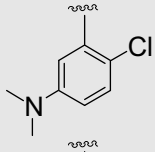
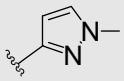
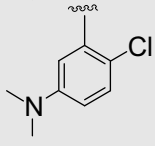
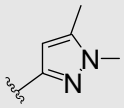
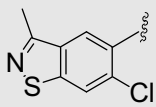
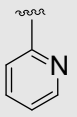
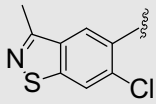
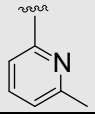


Compound	R^1	R^2	R^3	R^4	hOX_1R[a]	hOX_2R[a]
31	CN	H	H	Me_2N	166	10
32	Cl	H	H	Me_2N	119	5
33	Cl	H	H	CN	>10,000	463
34	Cl	H	H	CF_3	2486	48
35	OMe	H	H	Me_2N	>10,000	239
36	CN	Me_2N	H	H	>10,000	110
37	H	Me_2N	Cl	H	>10,000	789
38	CN	H	Me	H	886	4
39	Me_2N	H	H	Cl	4875	60
40	Cl	H	H	Me	1107	8
41	Cl	H	H	*i*-Pr	888	90
42	Cl	H	H	Et_2N	>10,000	2405
43	Cl	H	H	Me(Et)N	1373	433
44	Cl	H	H		2527	1606
45	Cl	H	H		1709	1161

[a] IC_{50} values in nM (FLIPR assay).

Table 3
Optimized orexin antagonists



Compound	R^1	R^2	hOX_1R[a]	hOX_2R[a]
46		Me	29	4.5
47			17	5
4			5	3
49			12	7
50			6	4
51			8	5
52			8	7

[a] IC_{50} Values in nM (FLIPR assay).

42 and **43**) or cyclization (compounds **44** and **45**) resulted in a significant loss of potency on both orexin receptors.

To identify more potent dual antagonists and to improve physicochemical properties of these sulfonamide orexin antagonists, compound **32** was selected as the starting point for further optimization. These structural modifications consisted of introducing heteroaromatic ring systems in the anilinic and the tertiary amide regions (Table 3). Interestingly, replacement of the 2-chloro-5-dimethylamino-phenyl moiety by a 6-chloro-3-methyl-benzisothiazole group resulted in significantly better affinity towards the OX_1R (compound **46**). Introduction of polar heterocyclic systems in the tertiary amide region yielded the most potent dual orexin antagonists identified so far in this class (compounds **47**–**52**).

Pharmacokinetic studies with selected compounds were carried out in Wistar rats[8] at intravenous and oral doses of 1 and 10 mg/kg, respectively, (Tables 4 and 5) and demonstrated that encouraging profiles with significant exposure were achieved. Generally, the compounds exhibited low systemic plasma clearance except for compound 51 (51 mL/min kg), good oral bioavailability ($F > 30\%$) and high AUC (>2300 ng h/mL). Compound **49** had the best pharmacokinetic profile and possessed an oral bioavailability of 46%. Surprisingly, replacement of the 2-chloro-5-dimethylamino-phenyl moiety by the 6-chloro-3-methyl-benzisothiazole substituent, as in compounds **51** and **52**, resulted in a tremendous loss of the pharmacokinetic profile with an oral bioavailability as low as for compound 1.

In vivo effects on sleep and wake cycles and blood–brain barrier (BBB) penetration were evaluated for compounds **47**, **48**, and **50** in adult male Wistar rats.

Effects of those compounds were assessed in freely moving rats implanted with radiotelemetric probes recording electroencephalographic (EEG) and electromyographic (EMG) signals, as well as locomotor activity and body temperature. We have previously

Table 4
Pharmacokinetic parameters of compounds **47**–**52** in rats following intravenous administration at a dose of 1 mg/kg

Compound	$t_{1/2}$[a]	AUC[b]	Clp[c]	V_{ss}[d]
47	0.5	434	38	1.7
48	1.5	1290	13	1.5
49	0.9	1230	14	1
50	0.8	1140	15	0.7
51	0.6	327	51	1.0
52	0.8	536	32	1.5

[a] $t_{1/2}$ Apparent terminal half-life expressed in hours.
[b] AUC, area under the plasma concentration versus time curve expressed in ng h/mL.
[c] Clp, plasma clearance expressed in mL/min kg.
[d] Vss, volume of distribution at steady state expressed in L/kg.

Table 5
Pharmacokinetic parameters of compounds **47**–**52** in rats following oral administration at a dose of 10 mg/kg

Compound	C_{max}[a]	AUC[b]	T_{max}[c]	F[d]
47	582	2290	0.5	35
48	754	3860	1.0	30
49	858	5660	4.0	46
50	872	4570	0.5	40
51	41	34	0.5	1
52	40	121	0.5	2

[a] C_{max}, the maximum observed plasma concentration expressed in ng/mL.
[b] AUC, area under the plasma concentration versus time curve expressed in ng h/mL.
[c] t_{max}, time to reach the maximum observed plasma concentration expressed in hours.
[d] *F*, oral bioavailability expressed in percent of dose.

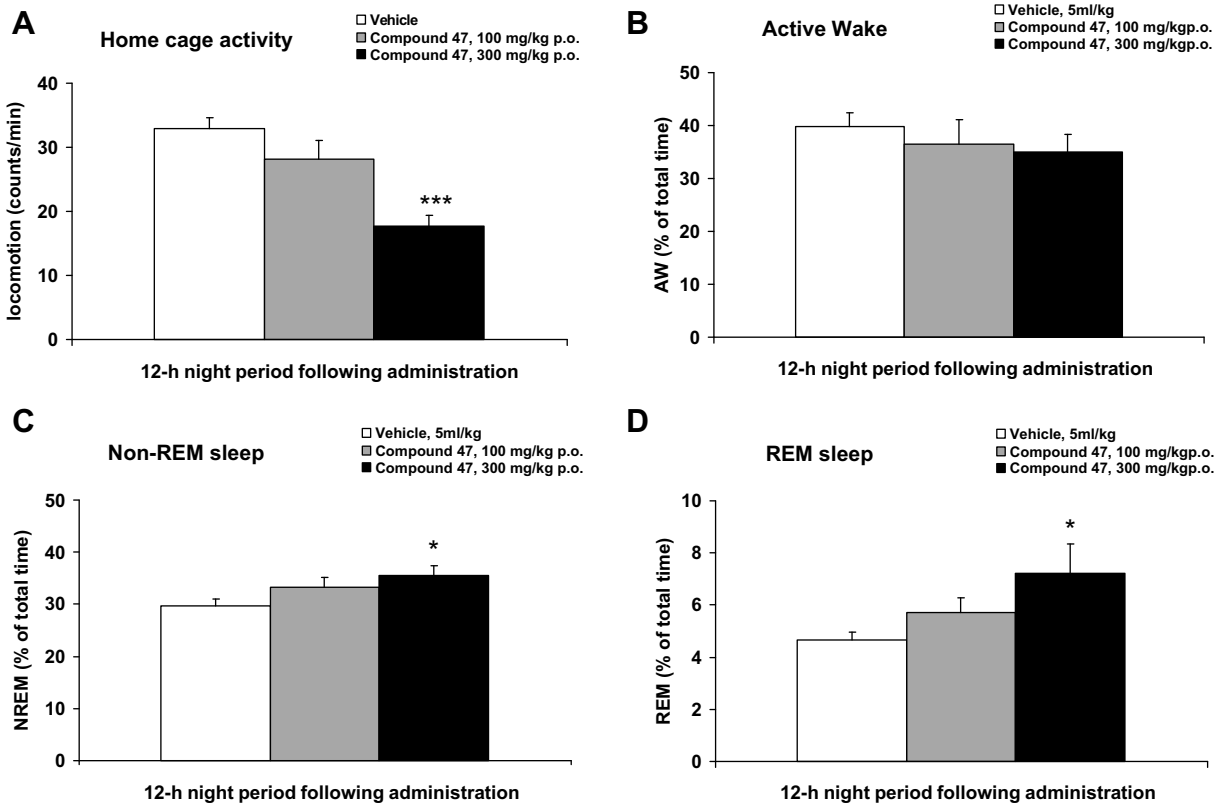


Figure 1. Dose–response effects of oral administration of compound **47**, a dual OX_1/OX_2 receptor antagonist. Effects on home cage activity (A), time spent in active wake (AW, B), time spent in non-REM sleep (NREM, C) and time spent in REM sleep (D) were measured and integrated over the 12-h night period following administration. Data are represented as means ± SEM. $^{*}p < 0.05$, $^{***}p < 0.001$ ($n = 15$ for vehicle, $n = 7$ for 100 mg/kg and $n = 8$ for 300 mg/kg).

shown that almorexant (or ACT-078573), a dual OX_1R/OX_2R antagonist that crosses the blood–brain barrier decreases alertness and increases non-REM and REM sleep in a dose-dependent manner when administered at the beginning of the dark active phase in rats.[5]

Tested under the same conditions (compound given orally at the beginning of the dark active phase), compound **47** was the only representative of this series that showed signs of activity on sleep and wake stages. It was tested at 100 and 300 mg/kg po and induced an apparent dose-dependent decrease in home cage activity ($P < 0.0001$, one-way ANOVA; Fig. 1A). The effect was significant at 300 mg/kg ($P < 0.0001$, Bonferroni post hoc analysis) when compared to vehicle control. We also observed a decrease in time spent in active wake over the 12 h night period which was not significant (($P > 0.05$, one-way ANOVA; Fig. 1B). Time spent in non-REM and REM sleep were both increased in a dose-dependent manner over the 12 h night period following administration ($P = 0.0318$ for non-REM sleep and $P = 0.0263$ for REM sleep, one-way ANOVA). The effect was significant at 300 mg/kg ($P < 0.05$, Bonferroni post hoc analysis, Fig. 1C and D) when compared to vehicle control. Body temperature was not significantly affected at any dose ($P > 0.05$, one-way ANOVA).

BBB penetration was evaluated at two different time points (1 and 3 h) following oral administration of 100 mg/kg. None of those compounds (**47**, **48**, and **50**) showed substantial total brain penetration. Concentration in brain was very low, in the range of 10–20 ng/g. Compound **47** showed the highest brain concentration observed for this class of compounds, reaching 66 ng/g 3 h following administration.

These total brain concentrations were sufficient to induce electrophysiological signs of non-REM and REM sleep when compound **47** was administered at three times higher dosage (300 mg/kg po, Fig. 1). Time spent in non-REM and REM sleep increased in physiological proportion: over the 12 h night period, non-REM sleep represented 86% of the total sleep time for vehicle-treated animals and 83% for animal treated with compound **47** (at 300 mg/kg) and REM sleep represented 14% of the total sleep time for the vehicle group and 17% for the compound **47** group. These pharmacological effects were achieved at total brain concentrations which are likely to be lower than those needed for almorexant to elicit similar effects on non-REM and REM sleep.[5] Characteristic physicochemical parameters (such as protein binding in plasma and brain or partitioning into lipid and aqueous compartments) are likely to contribute to differences in free extracellular concentrations reached in the brain for compounds of different classes.

Despite such differences, our investigations with these *N*-glycine-sulfonamides confirm the unique characteristics of dual orexin receptor antagonists. By transiently blocking the effects of endogenous orexin-peptides, those antagonists mimic the physiological state of sleep occurring when orexinergic neurons stop firing.[4]

In conclusion, we have described a novel series of potent dual orexin antagonists based on a *N*-glycine sulfonamide scaffold. SAR studies based on screening hit 1 have been outlined. A combination of a 2-chloro-5-dimethylamino-phenyl or a 6-chloro-3-methyl-benzisothiazole moiety in the anilinic region with polar heterocyclic systems in the tertiary amide region led to the most potent dual orexin receptor antagonists found so far in the literature. Generally, the series exhibited good oral bioavailability but was associated with low BBB penetration. We further demonstrated the ability of compound **47** to increase electrophysiological markers of both non-REM and REM sleep in rats following oral administration (at 300 mg/kg).

Acknowledgments

The authors thank K. Menyhart and C. Müller for carrying out the FLIPR assays, S. Delahaye, A. Weigel and C. Gnerre for performing the pharmacokinetic experiments, J. Hazemann, J. Gabillet, V. Egger, S. Paput, P. Rebmann, U. Fusco, D. Trachsel, D. Lehman, R. Haener, C. Fischer for expert technical support, T. Weller, C. Hubschwerlen, and M. Clozel for constantly supporting the project and Prof. H. Ramuz for stimulating discussions.

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.bmcl.2008.09.079.

References and notes

1. de Lecea, L.; Kilduff, T. S.; Peyron, C.; Gao, Y. X.; Foye, P. E.; Danielson, P. E.; Fukuhara, C.; Battenberg, E. L.; Gautvik, V. T.; Bartlett, F. S., 2nd; Frankel, W. N.; van den Pol, A. N.; Bloom, F. E.; Gautvik, K. M.; Sutcliffe, J. G. *Proc. Natl. Acad. Sci. U.S.A.* **1998**, *95*, 322.
2. Sakurai, T.; Amemiya, A.; Ishii, M.; Matsuzaki, I.; Chemelli, R. M.; Tanaka, H.; Williams, S. C.; Richardson, J. A.; Kozlowski, G. P.; Wilson, S.; Arch, J.; Buckingham, R. E.; Haynes, A. C.; Carr, S. A.; Annan, R. S.; McNulty, D. E.; Liu, W.; Terrett, J. A.; Elshourbagy, N. A.; Bergsma, D. J.; Yanagisawa, M. *Cell* **1998**, *92*, 573.
3. (a) Peyron, C.; Faraco, J.; Rogers, W.; Ripley, B.; Overeem, S.; Charnay, Y.; Nevsimalova, S.; Aldich, M.; Reynolds, D.; Albin, R.; Li, R.; Hungs, M.; Pedrazzoli, M.; Padigaru, M.; Kucherlapati, M.; Fan, J.; Maki, R.; Lammers, G. J.; Bouras, C.; Kucherlapati, R.; Nishino, S.; Mignot, E. *Nat. Med.* **2000**, *6*, 991; (b) Nishino, S.; Ripley, B.; Overeem, S.; Lammers, G. J.; Mignot, E. *Lancet* **2000**, *355*, 39.
4. (a) Lee, M. G.; Hassani, O. K.; Jones, B. E. *J. Neurosci.* **2005**, *25*, 6716; (b) Estabrooke, I. V.; McCarthy, M. T.; Ko, E.; Chou, T. C.; Chemelli, R. M.; Yanagisawa, M.; Saper, C. B.; Scammell, T. E. *J. Neurosci.* **2001**, *21*, 1656.
5. Brisbare-Roch, C.; Dingemanse, J.; Koberstein, R.; Hoever, P.; Aissaoui, H.; Flores, S.; Mueller, C.; Nayler, O.; van Gerven, J.; de Haas, S. L.; Hess, P.; Qiu, C.; Buchmann, S.; Schertz, M.; Weller, T.; Fischli, W.; Clozel, M.; Jenck, F. *Nat. Med.* **2007**, *13*, 150.
6. *FLIPR assay*. Chinese hamster ovary (CHO) cells expressing the human orexin receptors (hOX_1R or hOX_2R) were seeded into 96-well plates and incubated at 37 °C in 5% CO_2 with the cytoplasmic fluorescent calcium indicator fluo-3 AM (Molecular Probes). After washing the cells, intercellular Ca^{2+} mobilization was monitored as a change in cell fluorescence intensity by FLIPR (Molecular Devices). Differing concentrations of orexin antagonists were added to the plates prior to addition of orexin A. For each antagonist, IC_{50} (the concentration of compound needed to inhibit 50% of the agonistic response) is calculated.
7. Heffernan, G. D. U.S. Patent 6,562,979, 2003.
8. In vivo experiments were done on male Wistar rats. Rats are maintained on a 12-h light/12-h dark cycle.For pharmacokinetic studies, the formulation used for iv dosing was a 5%/95% (v/v) mixture of DMSO and 30/70 hydroxypropyl-*b*-cyclodextrine: sterile water and for po dosing was 100% PEG-400. Serial blood samples were collected into containers with Na-EDTA as anti-coagulant at various time-points and blood centrifuged to yield plasma. These studies used three animals (po) and one or two animals (iv).For BBB penetration studies, we measured the concentration of the OX receptor antagonist in plasma, CSF and brain sampled 1 and 3 h following oral administration. Plasma and brain are collected from the same animal at the same time (±5 min). Blood is sampled from the vena cava caudalis into containers with EDTA as anticoagulant and centrifuged to yield plasma. CSF is sampled from the cisterna magna with careful attention to avoid blood contamination. Brain is sampled after cardiac perfusion of 10 mL NaCl 0.9% and homogenized into 1 volume of cold phosphate buffer (pH 7.4).
For pharmacodynamic sleep studies, animals were implanted with miniature radiotelemetric implants (Data Sciences International) under general anesthesia. Those implants consist of two pair of differential leads; one pair for cranial placement to record the electroencephalogram (EEG) and one pair placed in either side of the muscular neck to record the electromyogram (EMG). This technology allows stress-free acquisition of vigilance and sleep stages, spontaneous activity and body temperature from freely moving rats in their home cage environment. Compounds were administrated orally at the beginning of the night dark cycle and formulated in 100% PEG-400. In each experiment, we used groups of 7 or 8 rats, in a crossover design, with at least 4-day washout periods separating consecutive administrations.